

GENENTECH 030R1C5

PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant : Goddard, et al. (as amended)
Appl. No. : 10/036,342
Filed : December 26, 2001
For : POLYPEPTIDES THAT INDUCE CELL
PROLIFERATION (as amended)
Examiner : Daniel E. Kolker
Group Art Unit : 1646

DECLARATION UNDER 37 C.F.R. §1.808

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Dear Sir:

I hereby aver that the nucleic acid of SEQ ID NO: 56, which encodes the protein of SEQ ID NO: 57, was deposited with the American Type Culture Collection (ATCC) April 20, 1999 and was given ATCC deposit number 203948. Accordingly, the deposited material has been accepted for deposit under the Budapest Treaty on the International Recognition of the deposit of Microorganisms for the Purposes of Patent Procedure and all restrictions on the availability to the public of the material so deposited will be irrevocably removed upon granting of the patent. The deposit will be maintained for a term of at least 30 years and at least five (5) years after the most recent request for the furnishing of a sample of the deposit was received by the depository

The deposited material is identical to the biological material and was in the Applicant's possession at the time the application was filed.

GENENTECH, INC.

Date:

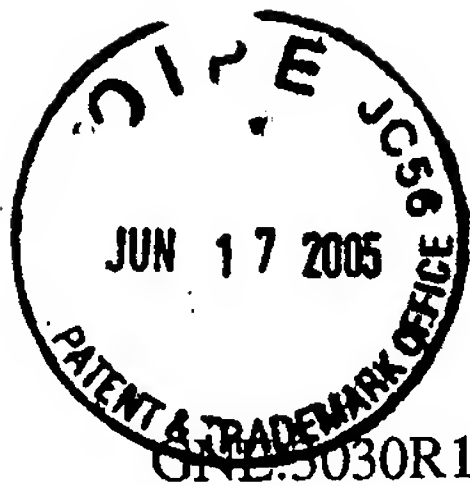
5/26/05

By:

[Signature]

Title:

PATENT AGENT



ONE:5030R1C5

PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant	:	Goddard et al. (as amended)
Appl. No.	:	10/036,342
Filed	:	December 26, 2001
For	:	POLYPEPTIDES THAT INDUCE CELL PROLIFERATION (as amended)
Examiner	:	Kolker, Daniel E.
Group Art Unit	:	1646

DECLARATION UNDER 37 CFR §1.131

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Dear Sir:

We declare and state as follows:

1. We are the inventors of the invention claimed in the above-captioned patent application.
2. During the time period in which we participated in the events and activities described herein, we were employed by Genentech, Inc., the assignee of the above-captioned application.
3. All of the events and activities described herein were performed by us personally, or by others at our direction as part of our duties as employees of Genentech, Inc.
4. The invention claimed in the above-captioned patent application was conceived and reduced to practice in the United States prior to November 18, 1999 as described below.
5. Prior to November 18, 1999, we conceived of the invention claimed in the above-captioned patent application. This is demonstrated by the attached sequence printout (Exhibit A), which was generated prior to November 18, 1999, and which shows the complete sequence of the nucleic acid having the sequence of SEQ ID NO: 56. The attached printout also shows the complete sequence of the polypeptide which has the sequence of SEQ ID NO: 57. As evidenced by the sequence printout, we were in possession of the complete nucleic acid and amino acid sequences prior to November 18, 1999.
6. The date deleted from Exhibit A is prior to November 18, 1999. This date was redacted pursuant to M.P.E.P. § 715.07. The date that remains is the date the report was printed, April 28, 2005.
7. After these initial experiments, we diligently reduced the claimed subject matter to practice by working to express and purify the encoded polypeptide and to run it systematically

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through many assays. The cDNA was deposited with the American Type Culture Collection (ATCC) on April 20, 1999 and assigned ATCC no. 203948. The protein of interest was assigned a "protein inventory number" (e.g., PIN1205-1), and this protein is a polypeptide having the sequence of SEQ ID NO:57, and is encoded by SEQ ID NO: 56.

8. Exhibit B shows that the protein lot designated PIN1205-1 was delivered to James Pan on a date prior to November 18, 1999 in order to perform assay ASY92, called "Mouse Mesangial Cell proliferation Assay." Also, as shown in Exhibit B, the assay was completed on a date prior to November 18, 1999. Exhibit B also shows that the tested polypeptides tested positive ("All Positives"), thereby confirming the ability of the encoded polypeptide to induce mesangial cell proliferation. Thus, actual reduction to practice occurred on a date prior to November 18, 1999.

9. The dates deleted from Exhibit B all are prior to November 18, 1999. These dates were redacted pursuant to M.P.E.P. § 715.07. The date that remains is the date the report was printed, April 28, 2005.

10. After reducing the invention to practice, we worked with the Genentech, Inc. patent department to prepare a non-provisional patent application, which included the sequences of SEQ ID NO:56 and SEQ ID NO:57, as well as the data showing the ability to induce mesangial cell proliferation. That application was filed on March 1, 2000 as PCT/US00/05601.

11. We hereby declare that all statements made herein of our own knowledge are true and that all statements made on information or belief are believed to be true, and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful statements may jeopardize the validity of the application or any patent issued thereon.

By: <u></u>	Date: <u>6/7/05</u>
Audrey Goddard	
By: _____	Date: _____
Paul J. Godowski	
By: _____	Date: _____
Austin L. Gurney	
By: _____	Date: _____
James Pan	
By: _____	Date: _____
Colin K. Watanabe	
By: _____	Date: _____
William I. Wood	

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By: _____

Audrey Goddard

Date: _____

By: _____

Paul J. Godowski

Date: 5/31/05

By: _____

Austin L. Gurney

Date: _____

By: _____

James Pan

Date: _____

By: _____

Colin K. Watanabe

Date: _____

By: _____

William I. Wood

Date: _____

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By: _____
Audrey Goddard

Date: _____

By: _____
Paul J. Godowski

Date: _____

By: _____
Austin L. Gurney

Date: 6/8/05

By: _____
James Pan

Date: _____

By: _____
Colin K. Watanabe

Date: _____

By: _____
William I. Wood

Date: _____

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By: _____
Audrey Goddard

Date: _____

By: _____
Paul J. Godowski

Date: _____

By: _____
Austin L. Gurney

Date: _____

By: _____
James Pan

Date: June 9/05

By: _____
Colin K. Watanabe

Date: _____

By: _____
William I. Wood

Date: _____

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Date: _____

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James Pan

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Colin K. Watanabe

Date: 6/8/2005

By: _____
William I. Wood

Date: _____

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Date: _____

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Paul J. Godowski

Date: _____

By: _____
Austin L. Gurney

Date: _____

By: _____
James Pan

Date: _____

By: _____
Colin K. Watanabe

Date: _____

By: _____
William I. Wood

Date: 5/21/05

EXHIBIT A

GENE GENES

GENEVIEWER

SEQUENCE VIEWER

ASSAY VIEWER

GENE

SEQ

PRO

TAP

SEC

BCU

MAP

RNA

EXP

GENEVIEW

LIB

PUR

LIB

PUR

LOT

LIB

PUR

LOT

LIB

PUR

LOT

Find

C

New

C

Update

ASY

92

66

Additional Resources

ASY92

Assay Name	Mouse Mesengial Cell proliferation Assay									
Alias Name	Mu Mess Cell Prolif									
Status	Retired									
Class	Primary									
Format	96 Well									
Type	Cell									
Sample										
Requirements										
Assay Volume	0.1 ml									
Fold Dil Into Well	10 Fold									
Replicates	3									
Dilutions	2									
Volume Requested	0.03ml/well/conc									
Protocol										
Species	Mouse									
Purpose	Screen SPDI proteins which can stimulate Mesengial Cell Proliferation									
Protocol	On day 1: Mouse mesengial cells are plated on a 96 well plate in Media[A 3:1 mbture of Dubecco's modified Eagle's medium and Ham's F12 medium- 95%- fetal bovine serum-5%- supplemented with 14mM hepes] and grw overnight. On day 2: SPDI Protehns are diluted at 2 conc. [1%- 0.1%] in serum-free Media and added to the cells. On day 4: After 48 hours incubation- each well of the plate was added 20 µl of the Cell Titer 96 Aqueous one solution reagent [Promega] and colormetric reaction was allowed for 2 hours. The absorbance [OD] is measured at 490 nm.									
Matrix	Promega kit for the assay-									
Result Calculation	replicated average									
Result Interpretation	Any PIN that gives an absorbance reading which is 15% above the media control is considered a hit.									
Result Cutoff	> 15 %									
In Vivo: InVitto:										
Comments										
Status	Retired									
Data Entered										
Data Canceled										
Department	Endocrinology									
Scientist	James (Guohua) Pan									
Notebook	0-									
Assayers										
Cancel Reason	Bioarea									
Lab Scientist	Wekuang Mao									

ASY | DNA | DOM | EXP | EAM | ELS | LIB | LOT | MAP | OLI | PBR | PRO | PUR | RNA | SEC | UNQ | XPT | YSI
Assay Viewer | Sequence Viewer | Gene Viewer | Genes

Genes Feedback

GENE GENES

SEQUENCE VIEWER

SEQUENCE VIEWER

SEQUENCE VIEWER

Find C New C Update

SELECT

Additional Resources

SPDI Assays

Assay Viewer

- ASY1 Heart Neonatal Hypertrophy
- ASY2 Heart Adult Hypertrophy
- ASY3 Adipocyte Lipolysis
- ASY4 Adipocyte Lipogenesis
- ASY5 Hematopoiesis: stem cell proliferation
- ASY6 Hippocampal Neuron Survival
- ASY7 Retinal Neuron Survival (5-6 days cultur
- ASY8 Endothelial cell proliferation
- ASY9 Inhibition of VEGF stimulated endothelia
- ASY10 Endothelial degradation [induction of]
- ASY11 B cell IgE synthesis inhibition

Find Lots

ALL PIN
ALL DNA

Show Lots for:

PIN

Number:

1205

☐ Include UNQ Related Lots

Lots for Search

PIN1205-1

☒ All Positives ☐ Verified Positives ☐ Pending

Date Complete From

To

ASSAY RESULT LIST

ASY	ASY Name	PUR/EXP/DNA	LOT	LOT Name	Pos	Verified	Conc	Conc Unit	Mean	Ctrl	UNQ	Protein Name	Data Complete	Comment
ASY92	Mu-Mess Cell Prolif	PUR1715	LOT2447	PIN1205-1			0.10	%	1		UNQ1915	Human DPKL1915 IgG		
ASY92	Mu-Mess Cell Prolif	PUR1715	LOT2447	PIN1205-1			1.00	%	1		UNQ1915	Human DPKL1915 IgG		

Page 1 of 1

Rows 1 - 2 of 2

Select Page Page No. 1

ASY | DNA | RNA | EXP | EAM | ELS | LIB | LOT | MAP | OLI | PRB | PRO | PUR | RNA | SRC | UNQ | XET | YST
Assay Viewer | Sequence Viewer | Gene Viewer | GenesGames | SAGE

GenesGames Feedback

EXHIBIT B

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>Thursday, April 28, 2005
>DNA92234 [Full]
>887 Sites [All Sites]
> [DNA92234], sheldens
> Lib309
>Sequence confirmed by phredphrap

      thai
      nlaIII  snaBI
      sphI   fnuDII/mvnI      mnlI
      nspHI  bstUI tail      taqI
      taiI  nspI  bsh1236I      xhoI
      maeII/hpyCH4IV bsiWI/splI  tsp509I[M.ecoRI-]
      aluI   hinII/acyI cac8I  bsaAI  ecoRI  tliI
      tsp45I  sapi  ahaII/bsaHI  mluI  rsaI  hpy188I  smlI
      maeIII  mboII  aatII  cac8I  aflIII  maeII/hpyCH4IV  paer7I  hpy188I  aciI  bpmI/g
      hphI   sfci  earI/ksp632I hpy99I hpyCH4V csp6I  aluI  apoI  avaI[M.taqI-]  mnlI  fnu4HI/bsoFI hpy18
1 TAGGTGACAC TATAGAAGAG CTATGACGTC GCATGCACGC GTACGTAAGC TCGGAATTTCG GCTCGAGGAA TGAATACCTC CGAAGCCGCT TTGTTCTCCA
  ATCCACTGTG ATATCTTCTC GATACTGCAG CGTACGTGGC CATGCATTTC AGCCTTAAGC CGAGCTCCTT ACTTATGGAG GCTTCGCGCA AACAAGAGGT
                                     ^insert starts here

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scrFI[M.hpaiI-]
nciI
mspI
hpaII
dsav
bpuAI bssKI bsp1286
bbsI bsII bsmFI taiI bmyI
aluI mboII bsaJI maeII/hpyCH4IV maeI maeIII nla
101 GATGTGAATA GCTCCACTAT ACCAGCCTCG TCTTCCTTCC GGGGGACAAC GTGGGTCAGG GCACAGAGAG ATATTTAATG TCACCTCTT GGGGCTTTCA
CTACACTTAT CGAGGTGATA TGGTCGGAGC AGAAGGAAGG CCCCCTGTTG CACCCAGTCC CGTGCTCTCTC TATAAATTAC AGTGGGAGAA CCCCCGAAAGT
sau3AI
mboI/ndeII[dam-]
dpnII[dam-]
dpnI[dam+]
alwI[dam-]
nlaIV
pleI mnlI bstYI/xhoII hgaI
mlyI rmaI bamHI bsII tseI
hinFI maeI hpy188I bstXI alwI[dam-] hpy188III fnu4HI/bsaI
bsmFI mnlI bfaI eco57I bpmI/gsuI[dcM-] bsII avai bbsI bsmFI
201 TGGGACTCCC TCTGCCACAT TTTTGGAGG TTGGGAAAGT TGCTAGAGGC TTCAGAACTC CAGCCTAATG GATCCCAAAC TCGGGAGAAT GGCTGCGTCC
ACCCTGAGG AGACGGTGTA AAAAACCTCC AACCTTTCA ACGATCTCCG AAGTCTTGAG GTCGGATTAC CTAGGGTTTG AGCCCTCTTA CCGACGCAGG
M D P K L G R M A A S
^MET
1

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fnu4HI/bsoFI
tseI aciI
tseI mwoI thai nlaIII haeII
mwoI fnu4HI/bsoFI nspHI mspI
fnu4HI/bsoFI fnuDII/mvnI scrFI[M.hpaII-]
bbvI bbvI bstUI[M.hhaI-] nciI
tseI tseI bsh1236I dsav hinPI bpuAI ms
mwoI fnu4HI/bsoFI hinPI nspI hphI mwoI hpaII bbsI rsaI mnII
fnu4HI/bsoFI hhaI/cfoI mnII aciI bssKI xmnI mboII csp6I econI
cac8I bbvI bpmI/gsuI[dcM-] bseRI mnII bsII bsaJI hhaI/cfoI asp700 bsri bsII
301 CTGCTGGCTG TGCTGCTGCT GCTGCTGGAG CGCGGCATGT TCCTCTCACC CTCCTCCGCC CCGGCGCTGT TAGAGAAAGT CTTCACAGTAC ATTGACCTCC
GACGACCGAC ACGACGACGA CGACGACCTC GCGCCGTACA AGAGGAGTGG GAGGGGCGGG GCGCGCGACA ATCTCTTTCA GAAGGTCATG TAACTGGAGG
12 L L A V L L L L L E R G M F S S P S P P P A L L E K V F Q Y I D L H

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mboII
earI/ksp632I
sapi
aluI
sstI
sacI
hgiAI/aspHI[M.aluI-
ecI136II
bsp1286[M.aluI-]
bsiHKAI hpy188I
hpy188I bmyI eco57I ea
banII[M.aluI-] cf
401 ATCAGGATGA ATTTGTGCAG ACGCTGAAGG AGTGGGTGGC CATCGAGAGC GACTCTGTCC AGCCTGTGCC TCGCTTCAGA CAAGAGCTCT TCAGAAATGAT
TAGTCCTACT TAAACACGTC TCGGACTTCC TCACCCACCG GTAGCTCTCG CTGAGACAGG AGCGAACGG AGCGAACGG GTTCTCGAGA AGTCTTACTA
46 Q D E F V Q T L K E W V A I E S D S V Q P V P R F R Q E L F R M M

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eaeI[dcM-]
scrFI[dcM-]
pspGI
mvaI

ecoRII[dcM-]
dsaV[dcM-]
bstNI bslI
bssKI[dcM-]
apyI[dcM+]
fokI cfrI bsrI
bstF5I haeIII/palI

601 CCCGTCATCC TGGCCGAACT GGGAGCGAT CCCACGAAAG GCACCGTGTG CTTCTACGGC CACTTGGACG TGCAGCCTGC TGACCGGGGC GATGGGTGGC
GGGCAGTAGG ACCGGCTTGA CCCCTCGCTA GGTGCTTTC CGTGGCACAC GAAGATGCCG GTGAACCTGC ACGTCGGACG ACTGGCCCCG CTACCCACCG
112 P V I L A E L G S D P T K G T V C F Y G H L D V Q P A D R G D G W L

scrFI[M.hpaII-]
ncII
tseI
fnu4HI/bsoFI mspI
haeIII/palI
bsgI cac8I hpaII
taiI bbvI
dsav
maeII/hpyCH4IV
bssKI
btrI hpyCH4V
bsaJI

sau3AI mwoI
bslI
sau96I[M.haeIII-] dpnII[dam-] hi
nlaIV
avaiI
accI
mnII
mcrI
bseRI bsiEI
eco0109I/draII
alwI[dam-] hae

701 TCACGGACCC CTATGTGCTG ACGGAGGTAG ACGGGAACCT TTATGGACGA GGAGCGACCG ACAACAAAGG CCTGTCTTG GCTTGGATCA ATGCTGTGAG
AGTGCCTGGG GATACACGAC TGCCTCCATC TGCCCTTTGA AATACCTGCT CCTCGCTGGC TGTGTTTCC GGGACAGAAC CGAACCTAGT TACGACACTC

146 T D P Y V L T E V D G K L Y G R G A T D N K G P V L A W I N A V S

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scrFI[dcm-]          mnII
pspGI                bpmI/gsuI[dcm-]
mvaI                  scrFI[dcm-]
ecoRII[dcm-]         pspGI
dsaV[dcm-]           mvaI
bstNI                 ecoRII[dcm-]
bspl1286              dsaV[dcm-]
bmyI bssKI[dcm-]     bstNI
hpy188I apyI[dcm+]   bstNI
eco57I bsaJI         bssKI[dcm-]
mwoI banII bpmI/gsuI[dcm-] apyI[dcm+]
                        foki mboII cac8I
                        bstF5I mnII
                        tsp509I
                        apoI mnII earI/ksp632I
                        bsaJI
801 CGCCTTCAGA GCCCTGGAGC AAGATCTTCC TGTGAATATC AAATTCAATCA TTGAGGGGAT GGAAGAGGCT GGCTCTGTG CCTGGAGGA ACTGTGGAA
    GCGGAAGTCT CGGGACCTCG TTCTAGAAGG ACACTTATAG TTTAAGTAGT AACTCCCCCTA CCTTCTCCGA CCGAGACAAC GGGACCTCCT TGAACACCTT
179 A F R A L E Q D L P V N I K F I I E G M E E A G S V A L E E L V E

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scrFI[
nciI
mspI
hpaII
dsav
bssKI
bsaJI
xmaI/ps
smaI
scrFI[M
nciI
dsav
bssKI
bsaJI
avaI[M.
nlaIV
901 AAAGAAAAGG ACCGATTCTT CTCTGGTGTG GACTACATTG TAATTCAGA TAACCTGTGG ATCAGCCAAA GGAAGCCAGC AATCACTTAT GGAACCCGGG
TTTCTTTTCC TGGCTAAGAA GAGACCACAC CTGATGTAAC ATTAAAGTCT ATTGACACACC TAGTCGGTTT CCTTCGGTCG TTAGTGAATA CCTTGGGCCC
212 K E K D R F F S G V D Y I V I S D N L W I S Q R K P A I T Y G T R G
tfII
sau96I mboII
avaII hinFI
sau3AI
mboI/ndeII[dam-]
dpnII[dam-]
dnpI[dam+]
hpy188I
tsp509I
alwI[dam-]
cac8I
nlaIV
901 AAAGAAAAGG ACCGATTCTT CTCTGGTGTG GACTACATTG TAATTCAGA TAACCTGTGG ATCAGCCAAA GGAAGCCAGC AATCACTTAT GGAACCCGGG
TTTCTTTTCC TGGCTAAGAA GAGACCACAC CTGATGTAAC ATTAAAGTCT ATTGACACACC TAGTCGGTTT CCTTCGGTCG TTAGTGAATA CCTTGGGCCC
212 K E K D R F F S G V D Y I V I S D N L W I S Q R K P A I T Y G T R G

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scrFI[dcn-]
pspGI
mvaI
ecorII[dcn-]
dsaV[dcn-]
bstNI
bssKI[dcn-]
bsmAI
bsaI
hphI
nlaIII
mnI
hpyCH4V
apyI[dcn+]
bspCNI
dclI
nlaIV
hpy188III
fokI
rcaI
nlaIII
sau3AI
sap
mboI/ndeII[da
dpnII[dam-]
dpnI[dam+] ea
aluI
nlaIII
mnI
hpyCH4V
apyI[dcn+]
bspCNI
dclI
nlaIV
hpy188III
fokI
rcaI
nlaIII
sau3AI
sap
mboI/ndeII[da
dpnII[dam-]
dpnI[dam+] ea
1001 GGAACAGCTA CTTCATGGTG GAGGTGAAAT GCAGAGACCA GGATTTTCAC TCAGGAACCT TTGGTGGCAT CCTTCATGAA CCAATGGCTG ATCTGGTTGC
CCTTGTCGAT GAAGTACCAC CTCCACTTTA CGTCTCTGGT CCTAAAAGTG AGTCCTTGGG AACCAACCGTA GGAAGTACTT GGTACCGAC TAGACCAACG
246 N S Y F M V E V K C R D Q D F H S G T F G G I L H E P M A D L V A

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scrFI[dcm-]
pspGI
mvaI
ecorII[dcm-]
dsaV[dcm-]
bstNI
bssKI[dcm-]
sau96I[dcm-]
nlaIV
    accI
    scrFI[dcm-]
    pspGI pleI
    mvaI mlyI
    ecorII[dcm-]
    dsaV[dcm-]
    bstNI hinfI
    bssKI[dcm-]
        mboII
        apyI[dcm+]
1101 TCTTCTCGGT AGCCTGGTAG ACTCGTCTGG TCATATCCTG GTCCCTGGAA TCTATGATGA AGTGGTTCCT CTTACAGAAG AGGAAATATA TACATACAAA
    AGAAGAGCCA TCGGACCATC TGAGCAGACC AGTATAGGAC CAGGACCTT AGATACTACT TCACCAAGGA GAATGTCTTC TCCTTTATTT ATGTATGTTT
279 L L G S L V D S S G H I L V P G I Y D E V V P L T E E I N T Y K
    xmnI nlaIV
    asp700
    mnlI
    earI/ksp632I
    mboII
    mnlI
    mnlI

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rsal
csp6I
nlaIV
kpnI
bani
asp718
bpmI/gsuI[dcn
hpy188III
fokI hpy188III mboII hpaII fnu4HI/bsoFI tsp509I mnlI tfiI mnlI acc65I
bstF5I bfaI bfaI bsaWI bsvI bbvI apoI taqI ddeI bseRI hinfI hpyCH4V mnlI
1201 GCCATCCATC TAGACCTAGA AGAATACCGG AATAGCAGCC GGGTTGAGAA ATTCTGTTC GATACTAAGG AGGAGATTCT AATGCACCTC TGGAGGTACC
CGGTAGGTAG ATCTGGATCT TCTTATGGCC TTATCGTCGG CCCAACTCTT TAAAGACAAG CTATGATTCC TCCTCTAAGA TTACGTGGAG ACCTCCATGG
312 A I H L D L E E Y R N S S R V E K F L F D T K E E I L M H L W R Y P

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haeIII/palI
eaeI[dcn-]
cfrI
thaI
fnuDII/mvni scrFI[dcn-] scrFI[dcn-]
hinPI pspGI
mnlI bstUI[M.hhaI-] mvaI
sau3AI hhaI/cfoI ecoRII[dcn-]
mboI/ndeII[dam-][M.taqI-]
dpnII[dam-] dsav[dcn-] bstNI tsp509I bsm
dpmI[dam+] bstNI bssKI[dcn-] apoI rmaI
alwI[dam-] bsh1236I bssKI[dcn-] apyI[dcn+] xmnI maeI
nlaIII taqI[dam-] apyI[dcn+] bst4CI/hpyCH4III asp700 bfaI
1301 CATCTCTTTC TATTCATGGG ATCGAGGGCG CGTTTGATGA GCCTGGAACT AAAACAGTCA TACCTGGCCG AGTTATAGGA AAATTTTCAA TCCGTCTAGT
GTAGAGAAAG ATAAGTACCC TAGCTCCCGC GCAAACTACT CGGACCTTGA TTTTGTCACT ATGGACCGGC TCAATATCCT TTATAAAGTT AGGAGATCA
346 S L S I H G I E G A F D E P G T K T V I P G R V I G K F S I R L V

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nlaiII	tsp45I	pl
mslI	maeIII	ml
mnli mslI	hphI	hi
aciI	hpy188III	nlaiII
	mboII	
	xmnI	
	asp700	bstXI
1401 CCCTCACATG AATGTGCTG CGGTGGAAGA ACAGGTGACA CGACATCTTG AAGATGTGTT CTCCTCAAGA AATAGTTCCA ACAAGATGGT TGTTCCTCATG		
GGGAGTGTAC TTACACAGAC GCCACCTTTT TGTCCTACTGT GCTGTAGAAC TTCTACACAA GAGGTTTCT TTATCAAGGT TGTTCCTACCA ACAAAGGTAC		
379 P H M N V S A V E K Q V T R H L E D V F S K R N S S N K M V V S M		

	tspRI	
	hpy188I	sau
	mwoI	mbo
	sau3AI bst4CI/hpyCH4III	dpn
rmaI	tseI	dpn
maeI	fnu4HI/bsoFI	dpn
bfaI	bsrI	alw
	bsaJI hpyCH4V	
1501 ACTCTAGGAC TACACCCGTG GATTGCAAAAT ATTGATGACA CCCAGTATCT CGCAGCAAAA AGAGCGATCA GAACAGTGT TGGAAACAGAA CCAGATATGA		
TGAGATCCTG ATGTGGGCAC CTAACGTTTA TAACTACTGT GGGTCATAGA GCGTCGTTT TCTCGCTAGT CTTGTCACAA ACCTTGTCTT GGTCTATACT		
412 T L G L H P W I A N I D D T Q Y L A A K R A I R T V F G T E P D M I		


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sau3AI
mboI/ndeII[dam-]
dpnII[dam-]
fokI dpnI[dam+]
    bstF5I
        scrFI[M.hpaII-]
            nciI  alwI[dam-]
            mspI  nlaIV
            hpaII bstYI/xhoII
            dsav  bamHI
                tsp509I
                    muniI/mfeI
                        bssKI alwI[dam-]
                            mspAII/ndeII[dam-]
                                scrFI[dcm-]
                                    pspGI mboI/ndeII[dam-]
                                        mvaI  dpnII[dam-]
                                            ecoRII[dcm-]
                                                dsav[dcm-]
                                                    bstNI dpnI[dam+]
                                                        mspAII/nspBII
                                                            bssKI[dcm-]
                                                                tsp509I
                                                                    mwoI  aciI  aluI
                                                                        apyI[dcm+]
                                                                            muniI/mfeI
                                                                                mspAII/ndeII[dam-]
                                                                                    scrFI[dcm-]
                                                                                        pspGI mboI/ndeII[dam-]
                                                                                            mvaI  dpnII[dam-]
                                                                                                ecoRII[dcm-]
                                                                                                    dsav[dcm-]
                                                                                                        bstNI dpnI[dam+]
                                                                                                            mspAII/nspBII
                                                                                                                bssKI[dcm-]
                                                                                                                    tsp509I
                                                                                                                        mwoI  aciI  aluI
                                                                                                                            apyI[dcm+]
                                                                                                                                muniI/mfeI
                                                                                                                                    mspAII/ndeII[dam-]
                                                                                                                                        scrFI[dcm-]
                                                                                                                                            pspGI mboI/ndeII[dam-]
                                                                                                                                                mvaI  dpnII[dam-]
                                                                                                                                                    ecoRII[dcm-]
                                                                                                                                                        dsav[dcm-]
                                                                                                                                                            bstNI dpnI[dam+]
                                                                                                                                                                mspAII/nspBII
                                                                                                                                                                    bssKI[dcm-]
                                                                                                                                                                        tsp509I
                                                                                                                                                                            mwoI  aciI  aluI
                                                                                                                                                                                apyI[dcm+]
                                                                                                                                                                                    muniI/mfeI
                                                                                                                                                                                        mspAII/ndeII[dam-]
                                                                                                                                                                                            scrFI[dcm-]
                                                                                                                                                                                                pspGI mboI/ndeII[dam-]
                                                                                                                                                                                                    mvaI  dpnII[dam-]
                                                                                                                                                                                                        ecoRII[dcm-]
                                                                                                                                                                                                            dsav[dcm-]
                                                                                                                                                                                                                bstNI dpnI[dam+]
                                                                                                                                                                                                                    mspAII/nspBII
                                                                                                                                                                                                                        bssKI[dcm-]
                                                                                                                                                                                                                            tsp509I
                                                                                                                                                                                                                                mwoI  aciI  aluI
                                                                                                                                                                    1601 TCCGGGATGG ATCCACCATT CCAATTGCCA AAATGTTCCA GGATCGTC CACAAGAGCG TGGTGCTAAT TCCGCTGGA GCTGTTGATG ATGGAGAACA
                                                                                                                                                                                                                                    AGGCCCTACC TAGGTGGTAA GGTAAACGGT TTTACAAGGT CCTCTAGCAG GTGTTCTCGC ACCACGATTA AGGCGACCCCT CGACAACCTAC TACCTCTTGT
                                                                                                                                                                                                                                    446 R D G S T I P I A K M F Q E I V H K S V V L I P L G A V D D G E H

tru9I
tseI
    nlaIV
        fnu4HI/bsoFI
            mnII  tsp509I  bbvI  ddeI
                sau96I[M.haeIII-]
                    haeIII/palI aseI/asnI/vspI
                        1701 TTCGCAGAAT GAGAAATCA ACAGGTGGAA CTACATAGAG GGAACCAAT TATTGCTGC CTTTCTTCTTA GAGATGGCCC AGCTCCATTA ATCACAAGAA
                        AAGCGTCTTA CTCTTTTAGT TGTCCACCTT GATGTATCTC CCTTGGTTTA ATAAACGACG GAAAAAGAAAT CTCCTACCGGG TCGAGGTAAT TAGTGTCTT
                        479 S Q N E K I N R W N Y I E G T K L F A A F F L E M A Q L H O

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sau3AI
mboI/ndeII[dam-]
dpnII[dam-]
dpnI[dam+]
    hpy188I
sau3AI    tspRI
    hpy188I    alwI[dam-]
        rmaI    mboI/ndeII[dam-]    hphI
        maeI    dpnII[dam-]    tfiI    mnlI    foki    bfaI    foki    bstF5I    bstF5I    hpy188III    maeI    rsaI
        bfaI    dpnI[dam+]    hinfI[M.hphI-]    bstF5I    bstF5I
1801 CCTTCTAGTC TGATCTGATC CACTGACAGA TTCACCTCCC CCACATCCCT AGACAGGGAT GGAATGTAA TATCCAGAGA ATTTGGGTCT AGTATAGTAC
    GGAAGATCAG ACTAGACTAG GTGACTGTCT AAGTGGAGGG GGTGTAGGA TCTGTCCCTA CCTTACATTT ATAGGTCTCT TAAACCCAGA TCATATCATG

sau96I
sau3AI
    mboI/ndeII[dam-]
    dpnII[dam-]
    dpnI[dam+]
        hpy188III
        tru9I    mseI
        ahaIII/draI    ecoRV    alwI[dam-]    sspl
1901 ATTTTCCCTT CCATTATAAA TGTCTTGGGA TATCTGGATC AGTAATAAAA TATTTCAAAG GCACAGATGT TGAATGTTT TTAAGGTCCC CCACTGCACA
    TAAAAGGGAA GGTAAATTTT ACAGAACCCT ATAGACCTAG TCATTATTTT ATAAAGTTTC CGTGTCTACA ACCTTTACCA AATTCACAGG GGTGACGTGT

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scrFI[dcM-]
pspGI
mvaI
ecoRII[dcM-]
dsaV[dcM-]
bstNI
bssKI[dcM-]
apyI[dcM+]
bsII      tfII
hpyCH4V   bsaJI   hinFI
2001 CCTTCCTCAA GTCATAGCTG CTTGATTTC CCAAGTCCTG TGCAATAGCC CCAGGATTGG ATTCCCTCCA ACCTTTTAGC ATATCTCCAA
GGAAGGAGTT CAGTATCGAC GAACGTCGTT GAACTAAAGG GGTTCAGGAC ACGTTATCGG GGTCCTAACC TAAGGAAGGT TGGAAAAATCG TATAGAGGTT

tseI
cac8I
tseI   fnu4HI/bsoFI
fnu4HI/bsoFI
smlI   bbvI   bbvI
mnII   aluI   hpyCH4V
2101 CCTTCCTCAA GTCATAGCTG CTTGACTCCG GTTGTCTTC TAGTCTCCTCA AGTGCTCGTG ACACATAATC ATTCCATCCA ATGATCGCCT TTGCTTTACC
GGAACGTTAA ACTAACCGTA TTAGTGAGGC CAAACGAAAG ATCCAGGAGT TCACGAGCAC TGTGTATTAG TAAGGTAGGT TACTAGCGGA AACGAAATGG

sau96I   tsp45I
avaII    bssSI
ppuMI    hgiAI/aspHI
ecoO109I/draII hpy188III
rmaI     bsp1286
mspI     hpaII   smlI   bsiHKAI      foki
tsp509I  maeI     mnII   bmyI   maeIII   bstF5I   dpnII[dam-]
hpyCH4V  bfaI     bsaWI  bsaWI  bmyI   maeIII   dpnI[dam+]
2101 CCTTGCAATT TGATTGGCAT AATCACTCCG GTTGTCTTC TAGTCTCCTCA AGTGCTCGTG ACACATAATC ATTCCATCCA ATGATCGCCT TTGCTTTACC
GGAACGTTAA ACTAACCGTA TTAGTGAGGC CAAACGAAAG ATCCAGGAGT TCACGAGCAC TGTGTATTAG TAAGGTAGGT TACTAGCGGA AACGAAATGG

tru9I
mseI     bsmAI
aseI/asnI/vspI   bsaI   tspRI
2201 ACTCTTTTCTT TTTATCTTAT TAATAAAAAAT GTTGGTCTCC ACCACTGNCT CCCAAAAAAA AAAAAAAA AAAAAAAA AAAAAAAA
TGAGAAAGGA AAATAGAATA ATTATTTTTA CAACCAAGAGG TGGTGACNGA GGGTTTTTTT TTTTTTTTTT TTTTTTTTTT TTTTTTTTTT

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scrFI[M.hpaII-]
nciI
mspI
hpaII
dsav
bssKI          sau96I rsal
xmaI/pspAI     rsrII/cspI
smaI           mroI   nlaIV
          scrFI[M.hpaII-] cpoI kpnI hpyCH4V
          aciI
          fnu4HI/bsoFI      taqI nciI      hpy188III csp6I
          haeIII/pali      sstI salI dsav      bspMI   banI sfcI
          mcrI            sacI hincII/hindII[M.taql-] avaiI[M.hpaII-]
          eagI/xmaIII/eclXI aluI accI[M.taql-] tru9I mspI   asp718
          eaeI            hgiAI/aspHI[M.aluI-] msei bspEI cfr10I/bsrFI
          cfrI            rmaI ecl136II      bssKI aseI/asnI/vspI acc65I cac8I
          bsiEI           maeI bsp1286[M.aluI-] xmnI tsp509I bsaWI pstI
          notI           bfaI bsiHKAI      bsaJI tsp509I bsaWI ageI sse8387I
          fnu4HI/bsoFI     bmyI hpy99I avai[M.hpaII-] hpaII mspI bspMI   rsal
          aciI           speI banII[M.aluI-] asp700 accIII hpaII sbfi   csp6I aluI   sf
2301 AAAAAAAAAA AAGGGCGGC CGCGGACTAG TGAGCTCGTC GACCCGGGAA TTAATTCGG ACCGGTACCT GCAGGCGTAC CAGCTTTCCC
TTTTTTTTTT TTTTTCCTGATC ACTCGAGCAG CTGGGCCCTT AATTAAGGCC TGGCCATGGA GTCCCGCATG GTCGAAAGGG

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pleI
mlyI
hinfi          aluI
2401 TATAGTGAGT CGTATTAGAG CTGG
ATATCACTCA GCATAATCTC GAACC

```

> length: 2425

aatII (GACGTC):	25
acc65I (GGTACC):	1295 2374
accI (GTMKAC):	727 1117 2348
accIII (TCCGGA):	2366
acII (CCGC):	86 332 355 511 1420 1672 2326 2330
acyI (GRCGYC):	25
afIII (ACRYGT):	37
ageI (ACCGGT):	2371
ahaII (GRCGYC):	25
ahaIII (TTTAAA):	1914
aluI (AGCT):	19 48 110 485 569 1006 1680 1781 2016 2343 2392 2419
alw26I (CAGNNCTG):	418 523 565
alwI (GGATCNNNN):	270 271 628 785 959 1319 1599 1609 1610 1817 1936
alwNI (CAGNNCTG):	418 523 565
apaI (GGGCCC):	533
apoI (RAATTY):	54 409 841 1249 1381 1879
apyI (CCWGG):	528 609 813 882 1038 1113 1137 1144 1342 1363 1638 2061
aseI (ATTAAT):	1787 2219 2360
asni (ATTAAT):	1787 2219 2360
asp700 (GAANNNTTC):	375 1159 1379 1469 2358
asp718 (GGTACC):	1295 2374
asphi (WGCWC):	484 2152 2342
aspi (GACNNNGTC):	451
avaI (CYCGRG):	62 280 995 2353
avaII (GGWCC):	559 705 909 1140 1985 2143 2369
balI (TGGCCA):	437
bamHI (GGATCC):	270 1609
banI (GGYRCC):	640 1295 2374

banII (GRCYC) :	484 533 809 2342
bbsI (GAAGACNNNNN) :	130 379 587
bbvI (GCAGC) :	292 312 315 318 321 508 519 522 567 570 672 1235 1552 1756 2017 2024
bceAI (ACGGCNNNNNNNNNN) :	502 656
bfaI (CTAG) :	243 1210 1216 1396 1504 1805 1849 1889 2140 2337
bglI (GCCNNNNNGGC) :	535
bglII (AGATCT) :	822
bmyI (GDGCHC) :	159 484 533 809 2152 2342
bpmI (CTGGAG) :	96 258 325 814 883 1290
bpuAI (GAAGACNNNNNNN) :	130 379 587
bsaAI (YACGTR) :	42
bsaHI (GRCGYC) :	25
bsaI (GGTCTCNNNNN) :	1034 2234
bsaJI (CCNNGG) :	139 359 503 528 545 684 812 881 995 996 1143 1516 2060 2353
bsaWI (WCCGGW) :	1226 2127 2366 2371
bserI (GAGGAGNNNNNNNNN) :	342 749 1270
bsgI (GTGCAG) :	415 670 1994
bsh1236I (CGCG) :	38 331 1329
bsiEI (CGRYCG) :	755 2327
bsiHKAI (GWGCWC) :	484 2152 2342
bsiWI (CGTACG) :	40
bslI (CCNNNNNNNGG) :	135 184 274 275 354 396 614 631 771 1847 1848 2060
bsmAI (GTCTC) :	1034 2235
bsmAI (GTCTC) :	1034 2235
bsmFI (GGGACNNNNNNNNNNN) :	143 202 297 1141 1399 1986
bsoFI (GCNGC) :	85 292 312 315 318 321 332 508 519 522 567 570 672 1235 1552 1756
	2017 2024 2326 2329
bsp120I (GGGCCC) :	533
bsp1286 (GDGCHC) :	159 484 533 809 2152 2342
bspCNI (CTCAGNNNNNNNNN) :	563 1050

bspEI (TCCGGA) :	2366
bspHI (TCATGA) :	1074
bspMI (ACCTGC) :	2377
bspMII (TCCGGA) :	2366
bsrFI (RCCGGY) :	2371
bsrI (ACTGGN) :	384 618 1542
bssKI (CCNGG) :	139 360 528 609 684 813 882 995 996 1038 1113 1137 1144 1239 1342
	1363 1602 1638 2061 2353 2354
bssSI (CTCGTG) :	2155
bst4CI (ACNGT) :	643 1354 1573
bstAPI (GCANNNNNTGC) :	641
bstDSI (CCRYGG) :	503 1516
bstF5I (GGATG) :	405 606 857 1068 1203 1605 1844 1857 2175
bstNI (CCWGG) :	528 609 813 882 1038 1113 1137 1144 1342 1363 1638 2061
bstUI (CGCG) :	38 331 1329
bstXI (CCANNNNNTGG) :	260 1478
bstYI (RGATCY) :	270 822 1609
btgI (CCRYGG) :	503 1516
btri (CACGTC) :	667
btsI (GCAGTGN) :	1992
cac8I (GCNNGC) :	31 35 303 675 868 975 2020 2381
cfoI (GCGC) :	330 364 525 800 1328
cfr10I (RCCGGY) :	2371
cfrI (YGGCCR) :	437 500 611 657 1365 2327
cpoI (CGGWCCG) :	2368
csp6I (GTAC) :	41 387 1296 1897 2375 2387
cspI (CGGWCCG) :	2368
ddeI (CTNAG) :	563 1050 1265 1767
dpnI (GATC) :	271 628 786 823 960 1090 1320 1566 1599 1610 1644 1812 1817 1937
	2183

dpnII (GATC): 271 628 786 823 960 1090 1320 1566 1599 1610 1644 1812 1817 1937
 2183
 draI (TTTAA): 1914
 draII (RGNCCY): 532 558 768 1984 2142
 draIII (CACNNGTG): 642
 dsaI (CCRYGG): 503 1516
 dsaV (CCNGG): 139 360 528 609 684 813 882 995 996 1038 1113 1137 1144 1239 1342
 1363 1602 1638 2061 2353 2354
 eaeI (YGGCCR): 437 500 611 657 1365 2327
 eagI (CGGCCG): 2327
 earI (CTCTTCNNNN): 15 487 862 1100 1177
 ecl136II (GAGCTC): 484 2342
 eclXI (CGGCCG): 2327
 eco57I (CTGAAG): 250 424 474 489 804
 ecoNI (CCTNNNNNAGG): 396
 ecoO109I (RGNCCY): 532 558 768 1984 2142
 54
 ecorI (GAATTC): 528 609 813 882 1038 1113 1137 1144 1342 1363 1638 2061
 ecorII (CCWGG): 1929
 ecorV (GATATC): 85 292 312 315 318 321 332 508 519 522 567 570 672 1235 1552 1756
 fnu4HI (GCNGC): 2017 2024 2326 2329
 fnuDII (CGCG): 38 331 1329
 foki (GGATG): 405 606 857 1068 1203 1605 1844 1857 2175
 gsuI (CTGGAG): 96 258 325 814 883 1290
 haeII (RGGCY): 363 524 799
 haeIII (GGCC): 438 501 534 543 612 658 769 1366 1776 2328
 hgaI (GACGC): 295 420
 hgiAI (GWGCWC): 484 2152 2342
 hhaI (GCGC): 330 364 525 800 1328
 hinPI (GCGC): 330 364 525 800 1328

hincII (GTYRAC):	2348
hindII (GTYRAC):	2348
hinfI (GANTC):	204 451 585 914 1120 1148 1275 1500 1829 2070 2407
hinII (GRCGYC):	25
hpaII (CCGG):	139 361 684 996 1227 1239 1602 2128 2354 2367 2372
hphI (GGTGA):	3 181 346 1023 1434 1832
hpy188I (TCNGA):	51 79 252 476 491 582 806 946 1568 1809 1814
hpy188III (TCNNGA):	97 281 402 443 1051 1074 1209 1289 1446 1873 1933 2156 2366
hpy99I (CGWCG):	27 2347
hpyCH4III (ACNGT):	643 1354 1573
hpyCH4IV (ACGT):	26 43 149 668
hpyCH4V (TGCA):	34 416 521 671 1030 1283 1524 1995 2023 2051 2104 2380
kpnI (GGTACC):	1295 2374
ksp632I (CTCTTCNNNN):	15 487 862 1100 1177
maeI (CTAG):	243 1210 1216 1396 1504 1805 1849 1889 2140 2337
maeII (ACGT):	26 43 149 668
maeIII (GTNAC):	4 180 1435 2158
mboI (GATC):	271 628 786 823 960 1090 1320 1566 1599 1610 1644 1812 1817 1937
	2183
mboII (GAAGA):	15 131 380 488 588 825 862 917 1101 1177 1219 1450
mcrI (CGRYCG):	755 2327
mfeI (CAATTG):	1622
mluI (ACGGGT):	37
mlyI (GAGTCNNNNN):	204 451 585 1120 1500 2407
mnII (CCTC):	65 77 126 185 209 227 246 344 350 396 469 545 562 598 724 749 853
	865 886 1021 1168 1180 1270 1287 1293 1324 1402 1738 1835 2005 2146
mroI (TCCGGA):	2366
mscI (TGGCCA):	437
mseI (TTAA):	175 1788 1915 1981 2220 2361
mslI (CAYNNNRTG):	400 1405 1407

mspAI (CMGCKG) :	568 1672
mspi (CCGG) :	139 361 684 996 1227 1239 1602 2128 2354 2367 2372
munI (CAATTG) :	1622
mvaI (CCWGG) :	528 609 813 882 1038 1113 1137 1144 1342 1363 1638 2061
mvnI (CGCG) :	38 331 1329
mwoI (GCNNNNNNNGC) :	303 312 315 321 357 502 535 641 650 793 802 1555 1665
nciI (CCSGG) :	139 360 684 995 996 1239 1602 2353 2354
ndeII (GATC) :	271 628 786 823 960 1090 1320 1566 1599 1610 1644 1812 1817 1937
	2183
nlaIII (CATG) :	32 199 336 555 1014 1075 1315 1407 1497
nlaIV (GNNCC) :	270 532 533 558 640 705 991 1054 1140 1164 1295 1609 1741 1985 2374
notI (GCGGCCGC) :	2326
nspBII (CMGCKG) :	568 1672
nspHI (RCATGY) :	31 335
nspi (RCATGY) :	31 335
paer7I (CTCGAG) :	62
pali (GGCC) :	438 501 534 543 612 658 769 1366 1776 2328
pflFI (GACNNNGTC) :	451
pleI (GAGTCNNNN) :	204 451 585 1120 1500 2407
ppuMI (RGGWCCY) :	558 1984 2142
pshAI (GACNNNNNGTC) :	553
pspAI (CCCGGG) :	995 2353
pspGI (CCWGG) :	528 609 813 882 1038 1113 1137 1144 1342 1363 1638 2061
pspOMI (GGGCCC) :	533
pstI (CTGCAG) :	520 2379
pvuII (CAGCTG) :	568
rcaI (TCATGA) :	1074
rmaI (CTAG) :	243 1210 1216 1396 1504 1805 1849 1889 2140 2337
rsaI (GTAC) :	41 387 1296 1897 2375 2387
rsrII (CGGWCCG) :	2368

sacI (GAGCTC) :	484 2342
salI (GTCGAC) :	2348
sapI (GCTCTTCNNNN) :	15 486 1099
sau3AI (GATC) :	271 628 786 823 960 1090 1320 1566 1599 1610 1644 1812 1817 1937 2183
sau96I (GGNCC) :	533 534 559 705 769 909 1140 1776 1985 2143 2369
sbfI (CCTGCAGG) :	2378
scrFI (CCNGG) :	139 360 528 609 684 813 882 995 996 1038 1113 1137 1144 1239 1342 1363 1602 1638 2061 2353 2354
sfanI (GCATC) :	1067
sfcI (CTRYAG) :	10 520 2379 2400
sfiI (GGCCNNNNNGGCC) :	534
smaI (CCCGGG) :	995 2353
smlI (CTYRAG) :	62 2006 2147
snaBI (TACGTA) :	42
speI (ACTAGT) :	2336
sphI (GCATGC) :	31
splI (CGTACG) :	40
sse8387I (CCTGCAGG) :	2378
sspI (AATATT) :	1528 1949
sstI (GAGCTC) :	484 2342
taiI (ACGT) :	26 43 149 668
taqI (TCGA) :	63 443 1259 1322 2349
tfiI (GAWTC) :	914 1148 1275 1829 2070
thai (CGCG) :	38 331 1329
tliI (CTCGAG) :	62
tru9I (TTAA) :	175 1788 1915 1981 2220 2361
tseI (GCWGC) :	292 312 315 318 321 508 519 522 567 570 672 1235 1552 1756 2017 2024
tsp45I (GTSAC) :	4 180 1435 2158
tsp509I (AATT) :	55 410 842 942 1250 1382 1623 1668 1748 1880 2107 2359 2363

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GSeqEdit, DNA92234 [Full], page 23